ILS
Integrated
Laboratory
Systems, Inc.

P.O. Box 13501
Research Triangle Park
North Carolina 27709
919.544.5857
Fax: 919.544.5091

October 29, 1997

Dr. Larry G. Hart
Executive Secretary
P.O. Box 12233
Research Triangle Park, NC 27709

re: The Genotoxicity of Phenolphthalein

Dear Dr. Hart:

Due to other commitments, I am unable to attend the October 30th public meeting on the 9th Report on Carcinogens. However, as principal investigator for the project in which the phenolphthalein *p53* study was conducted, I would like to comment on the classification of phenolphthalein as a "genotoxic" or a "nongenotoxic" carcinogen.

In a number of *in vivo* mouse studies, phenolphthalein induced a significant increase in the frequency of micronucleated erythrocytes. Based on these results, phenolphthalein is classifiable as an *in vivo* "genotoxic" agent. From a kinetochore analysis of micronuclei in *p53* transgenic mice ingesting 12000 ppm phenolphthalein for 6 months (the only dose evaluated for kinetochore status), phenolphthalein is capable of inducing both structural and numerical chromosomal damage, with numerical damage occurring more frequently.

The thymic tumors induced by phenolphthalein in this study exhibited a loss of heterozygosity for the wild-type *p53* allele. This finding is consistent with **chromosome loss** or **deletions** large enough to encompass the portion of the gene (exons 2 through 6) used for the molecular analysis. Whether **either, neither, or both mechanism(s)** are involved in the induction of thymic tumors in mice (or any other tumors in mice and rats) requires additional research.

The lack of a significant increase in DNA damage in blood leukocytes or liver parenchymal cells of phenolphthalein-treated mice, evaluated using the single cell gel (SCG) assay, should not be construed as proof that phenolphthalein is not genotoxic. The positive micronuclei response occurred in proliferating erythropoietic cells. Thus, the lack of a significant increase in DNA damage in blood leukocytes and liver could reflect tissue-specific differences in response rather than providing insight into the mechanism(s) by which phenolphthalein is carcinogenic.

Thanks for this opportunity.

Sincerely yours,

Raymond Tice

Raymond R. Tice, Ph.D.

cc: TK Rao, President